Genomic DNA preparation from Yeast

Overview :
This protocol is generously provided by Derek McCusker, PhD Group Leader, CNRS IBGC - UMR 5095 (CNRS-UBS).

Procedure :

1. Scrape a toothpick of yeast cells from a freshly grown plate and dispense into 1 mL of water in a screw-top 1.5 ml tube. Spin down and remove the supernatant. You should use only a small glob of cells-approximately 10µL

2. Add about 200 µL glass beads (1 eppendorf tube cap full)

3. Add 200 µL lysis buffer:

    - 2% v/v triton X-100
    - 1% w/v SDS
    - 100 mM NaCl
    - 10 mM Tris-Cl pH 8.0
    - 1 mM EDTA

4. Add 200 µL phenol/chloroform

5. Vortex for 1 minute. There's no need to vortex too violently as the cells will be completely munched by the phenol/chloroform

6. Add 400µL TE, vortex again very briefly to mix everything up

7. Spin for 5 min in microcentrifuge and take the supernatant and transfer to a new tube, taking care to avoid the goop at the interface

8. Add an equal volume of chloroform and vortex for 1 minute

9. Spin for 5 min

10. Add 2 volumes of ethanol to the supernatant and spin 5 minutes in the microfuge. Remove supernatant. Spin again. Remove last few microliters of supernatant. Speedvac for 5 mins

11. Dissolve pellet in 50-100 µL H20, use a range of concentrations of DNA for PCR